

CLAIMS

What is claimed is:

1. An isolated complex comprising:
 - (a) a DEP-1 polypeptide that is capable of specific association with a DEP-1 substrate polypeptide; and
 - (b) a DEP-1 substrate polypeptide that is in specific association with the DEP-1 polypeptide.
2. The complex of claim 1 wherein the DEP-1 polypeptide is selected from the group consisting of:
 - (a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,
 - (b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,
 - (c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,
 - (d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,
 - (e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,
 - (f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,
 - (g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,

(h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3.

3. An isolated complex comprising:

(a) a DEP-1 polypeptide that is capable of specific dephosphorylation of a DEP-1 substrate polypeptide; and

(b) a DEP-1 substrate polypeptide that is in specific association with the DEP-1 polypeptide.

4. The isolated complex of either claim 1 or claim 3 wherein the DEP-1 substrate polypeptide is selected from the group consisting of

(a) a polypeptide which comprises the amino acid sequence set forth in any one of SEQ ID NOS:4-6,

(b) a polypeptide comprising the amino acid sequence set forth in SEQ ID NO:7 which comprises a transmembrane domain and a cytoplasmic domain of the polypeptide of (a),

- (c) at least one p120^{ctn} polypeptide comprising an amino acid sequence as set forth in any one of SEQ ID NOS:8-12, and
- (d) a Gab1 polypeptide comprising an amino acid sequence as set forth in SEQ ID NO: 13.

5. An isolated complex comprising a DEP-1 polypeptide in specific association with a polypeptide selected from the group consisting of

- (i) a plakoglobin polypeptide comprising an amino acid sequence as set forth in any one of SEQ ID NOS:14-15 and 22, and
- (ii) a beta-catenin polypeptide comprising an amino acid sequence as set forth in SEQ ID NO:16,

wherein the DEP-1 polypeptide is selected from the group consisting of:

- (a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,
- (b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,
- (c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,
- (d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,
- (e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,
- (f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,
- (g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,

(h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3.

6. A method of identifying an agent that alters interaction of a DEP-1 polypeptide with a DEP-1 substrate polypeptide, comprising:

(a) exposing, in the absence and presence of a candidate agent, a sample comprising a DEP-1 polypeptide and a DEP-1 substrate polypeptide to conditions sufficient for formation of a complex comprising the DEP-1 polypeptide in specific association with the DEP-1 substrate polypeptide; and

(b) comparing a first level of the complex that is formed in the absence of the candidate agent to a second level of the complex that is formed in the presence of the candidate agent, wherein an alteration in the second level relative to the first level indicates that the agent alters interaction between the DEP-1 polypeptide and the DEP-1 substrate polypeptide.

7. A method of identifying an agent that alters dephosphorylation by a DEP-1 polypeptide of a DEP-1 substrate polypeptide, comprising:

(a) exposing, in the absence and presence of a candidate agent, a sample comprising a DEP-1 polypeptide and a DEP-1 substrate polypeptide to conditions sufficient for (i) formation of a complex comprising the DEP-1 polypeptide in specific association with the DEP-1 substrate polypeptide and (ii) determination of dephosphorylation of the DEP-1 substrate polypeptide; and

(b) comparing a first level of DEP-1 substrate polypeptide dephosphorylation in the absence of the candidate agent to a second level of DEP-1 substrate polypeptide dephosphorylation in the presence of the candidate agent, wherein an alteration in the second level relative to the first level indicates that the agent alters dephosphorylation by the DEP-1 polypeptide of the DEP-1 substrate polypeptide.

8. The method of either claim 6 or claim 7 wherein the DEP-1 polypeptide is selected from the group consisting of:

(a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,

(b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,

(c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,

(d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,

(e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,

(g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,

(h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3.

9. The method of either claim 6 or claim 7 wherein the DEP-1 substrate polypeptide is selected from the group consisting of

(a) a polypeptide which comprises the amino acid sequence set forth in any one of SEQ ID NOS:4-6,

(b) a polypeptide comprising the amino acid sequence set forth in SEQ ID NO:7 which comprises a transmembrane domain and a cytoplasmic domain of the polypeptide of (a),

(c) at least one p120^{ctn} polypeptide comprising an amino acid sequence as set forth in any one of SEQ ID NOS:8-12, and

(d) a Gab1 polypeptide comprising an amino acid sequence as set forth in SEQ ID NO: 13.

10. A recombinant expression construct comprising a regulated promoter operably linked to a polynucleotide encoding a DEP-1 polypeptide.

11. The recombinant expression construct of claim 10 wherein the regulated promoter is an inducible promoter.

12. The recombinant expression construct of claim 10 wherein the regulated promoter is a tightly regulated promoter.

13. The recombinant expression construct of claim 10 wherein the DEP-1 polypeptide is selected from the group consisting of:

(a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,

(b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,

(c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,

(d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,

(e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,

(g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,

(h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3.

14. A host cell comprising the recombinant expression construct of any one of claims 10-13.

15. A cell line derived from the host cell of claim 14.

16. An immortal cell line according to claim 15.

17. A cell line according to claim 16 wherein the host cell is selected from a cancer cell, a transformed cell and a malignant cell.

18. A method of altering transduction of a biological signal in a cell, comprising:

introducing into a cell a DEP-1 polypeptide that is capable of specific association with a DEP-1 substrate polypeptide under conditions and for a time sufficient to permit formation of a complex comprising the DEP-1 polypeptide in specific association with the DEP-1 substrate polypeptide, wherein:

- (i) the DEP-1 polypeptide is selected from the group consisting of:
 - (a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,
 - (b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,
 - (c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,
 - (d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,
 - (e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,
 - (f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,
 - (g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,
 - (h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3, and wherein

(ii) the cell comprises a DEP-1 substrate polypeptide that is selected from the group consisting of

(a) a polypeptide which comprises the amino acid sequence set forth in any one of SEQ ID NOS:4-6,

(b) a polypeptide comprising the amino acid sequence set forth in SEQ ID NO:7 which comprises a transmembrane domain and a cytoplasmic domain of the polypeptide of (a),

(c) at least one p120^{ctn} polypeptide comprising an amino acid sequence as set forth in any one of SEQ ID NOS:8-12, and

(d) a Gab1 polypeptide comprising an amino acid sequence as set forth in SEQ ID NO: 13.

19. The method of claim 18 wherein the step of introducing comprises inducing expression of a polynucleotide that encodes the DEP-1 polypeptide, wherein said polynucleotide is present within the cell.

20. The method of claim 18 wherein the step of introducing comprises transforming or transfecting the cell with a recombinant expression construct that comprises a polynucleotide which encodes the DEP-1 polypeptide.

21. A method of altering transduction of a biological signal in a cell, comprising contacting a cell with an agent,

(i) wherein the cell comprises a DEP-1 polypeptide and a DEP-1 substrate polypeptide, said DEP-1 polypeptide being capable of specific association with the DEP-1 substrate polypeptide to form a complex,

(ii) wherein the agent is capable of altering the specific association of the DEP-1 polypeptide with the DEP-1 substrate polypeptide,

(iii) wherein the DEP-1 polypeptide is selected from the group consisting of:

(a) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:2,

(b) a polypeptide which comprises the amino acid sequence set forth in SEQ ID NO:3,

(c) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a nucleotide sequence that is a reverse complement of SEQ ID NO:1,

(d) a truncated DEP-1 polypeptide which comprises at least the amino acid sequence set forth at positions 1205-1245 of SEQ ID NO:2, or a variant thereof,

(e) a mutant polypeptide which comprises at least one amino acid substitution in the amino acid sequence set forth in SEQ ID NO:2, wherein the amino acid substitution is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(f) a mutant polypeptide according to (e) wherein aspartate at position 1205 is substituted with alanine,

(g) a mutant polypeptide according to (e) wherein cysteine at position 1239 is substituted with serine,

(h) a mutant polypeptide which comprises an amino acid sequence as set forth at positions 997-1337 of SEQ ID NO:2, said mutant polypeptide comprising at least one amino acid substitution that is selected from a substitution of aspartate at position 1205 and a substitution of cysteine at position 1239,

(i) a mutant polypeptide according to (h) wherein aspartate at position 1205 is substituted with alanine,

(j) a mutant polypeptide according to (h) wherein cysteine at position 1239 is substituted with serine,

(k) a polypeptide that is encoded by a polynucleotide that hybridizes under moderately stringent conditions to a nucleic acid molecule which comprises a reverse complement of a nucleotide sequence that encodes a polypeptide selected from any one of (e)-(j),

(l) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:2, and

(m) a polypeptide to which binds an antibody that specifically recognizes a polypeptide that comprises the amino acid sequence set forth in SEQ ID NO:3, and

(iv) wherein the DEP-1 substrate polypeptide is selected from the group consisting of

(a) a polypeptide which comprises the amino acid sequence set forth in any one of SEQ ID NOS:4-6,

(b) a polypeptide comprising the amino acid sequence set forth in SEQ ID NO:7 which comprises a transmembrane domain and a cytoplasmic domain of the polypeptide of (a),

(c) at least one p120^{ctn} polypeptide comprising an amino acid sequence as set forth in any one of SEQ ID NOS:8-12, and

(d) a Gab1 polypeptide comprising an amino acid sequence as set forth in SEQ ID NO: 13.

22. The method of either claim 18 or claim 21 wherein formation of the complex results in dephosphorylation of the DEP-1 substrate polypeptide.

23. The method of claim 22 wherein the DEP-1 substrate polypeptide is selected from the group consisting of (i) a polypeptide which comprises the amino acid sequence set forth in any one of SEQ ID NOS:4-6, and (ii) a polypeptide comprising the amino acid sequence set forth in SEQ ID NO:7 which comprises a transmembrane domain and a cytoplasmic domain of the polypeptide of (i), and wherein at least one phosphorylated amino acid selected from the group consisting of the amino acid corresponding to position 1349 of SEQ ID NO:4 and the amino acid corresponding to position 1365 of SEQ ID NO:4 is dephosphorylated.

24. The method of either claim 18 or claim 21 wherein transduction of the biological signal results in altered cell proliferation, differentiation or survival.

25. The method of either claim 18 or claim 21 wherein transduction of the biological signal results in altered cellular morphogenesis or altered cellular motility.